## REMARKS

Reconsideration is respectfully requested. Claims 21-34 were previously pending in the application. Claims 21-34 have been amended without the addition of new matter. Accordingly, claims 21-34 are pending in the application.

### Claim Rejections – 35 U.S.C. § 101

Claims 21-34 stand rejected under 35 U.S.C. § 101 as allegedly being directed to non-statutory subject matter. The Examiner alleges that the scope of claim 21 is such that the claimed invention "encompasses a human embryonic cell wherein the progeny of the cell is a developed human being". Office Action, page 2. Accordingly, Applicants have amended claims 21-34 to delete "and progeny thereof" from these claims. Applicants respectfully submit that the claims as amended now cover isolated cells and subsequently isolated cells that result from cell division of these cells, thus excluding the possibility of progeny which is a human being since a human being does not read on isolated cells. (Emphasis added.)

### Claim Rejections – 35 U.S.C. § 102

Claims 21, 23-27, and 30-32 stand rejected inder 35 U.S.C. § 102(b) as allegedly being anticipated by Zhang et. al., JBC 270: 8501-8505 (1995) ("Zhang 1995"). The Examiner alleges among other things that the reference "teaches a HS-40 enhancer element (NF-E2/AP1-II) which comprises the nucleotide sequence of SEQ ID:1 (tctgagtca), see page 8503, Fig-1B, 3'NF-E2/AP1-II". Office Action, page 4. The Examiner further alleges that this reference teaches that mutant HS-40 enhancer comprising a 1-bp mutation in the 3'NF-E2/AP1 motif (gctgagtca to tctgagtca) exhibits "a 2-3 fold higher level of enhancer activity than did the wild type HS-40 enhancer (page 8502, col. 2, paragraph 6; page 8504, Fig. 3)". Office Action, page 4.

In response, Applicants submit that the Zhang 1995 reference only teaches transient transfection of the DNA constructs that are disclosed, not stable integration of the constructs into cellular chromosomes. In fact, the authors of the Zhang 1995 reference purposely used transient

transfection to avoid the confounding effects of position effects on gene expression that might result from chromosomal integration. The authors of the Zhang 1995 reference state, "since only transient expression assay has been used, our data most likely reflect the structure-function relation . . .". Page 8503, first column, first paragraph under the heading "Discussion". (Emphasis added.) In contrast, the present invention claims, in part, "at least one copy of a chromosomally integrated transgene" in independent claim 21. It is well known that DNA which is transiently transfected in mammalian cells, as was taught in the Zhang 1995 reference, does not undergo chromosomal integration. Thus, at least this element of claim 21 (and claims that depend from claim 21) is not taught by the Zhang 1995 reference.

In order to anticipate a claim under 35 U.S.C. § 102(b), a reference must teach each and every element of a claimed invention. MPEP 2131. As discussed above, the Zhang 1995 reference fails to teach each and ever element of the claimed invention. Thus, this requirement for maintaining a rejection under 35 U.S.C. § 102(b) is not met by the Zhang 1995 reference, and Applicants respectfully request that this ground for rejection be withdrawn.

**Claim Rejections – 35 U.S.C. § 103**

Claims 22, 28-29, and 33-34 stand rejected under 35 U.S.C. § 103(a) as allegedly being unpatentable over Zhang 1995, as applied to claims 21-27 and 30-32, and further in view of Zhang et al., Mol Cell Biol., 4:2298-308, 1993 ("Zhang 1993"). In making this rejection, the Examiner alleges that while Zhang 1995 teaches "a mutated HS-40 enhancer element (NF-E2/AP1-II) comprising the nucleotide sequence of SEQ ID NO:1 (tctgagtca), Zhang does not teach nucleic acid sequences comprising SEQ ID NO:2 and SEQ ID NO:3 of the instant application." Office Action, page 5.

The Examiner then cites the Zhang 1993 reference as disclosing a nucleotide sequence for the HS-40 enhancer element which matches the nucleotide sequences of SEQ ID NO:2 and SEQ ID:3 (page 2299, Fig. 1B). The Examiner points out that Zhang 1993 discloses SEQ ID NO:1 (TCTGAGTCA) on page 2304, column 1, Figure 7A of the Zhang 1993 reference. The Examiner

argues that it would be obvious to use flanking regions around the tctgagtca element in order to regulate erythroid lineage development in a stage-specific fashion. Office Action, page 5.

Applicants respectfully traverse these grounds for rejection. In order to establish a *prima facie* case of obviousness, an examiner must meet three basic criteria: (1) there must be some suggestion or motivation to modify a reference or combine reference teachings, (2) there must be a reasonable expectation of success, (3) the references must teach or suggest all claim limitations. MPEP 2142.

The combination of Zhang 1995 and Zhang 1993 fails to meet these criteria. First, the references alone or in combination fail to teach or suggest all claim limitations. Independent claim 21 recites as one of its limitations "at least one copy of a chromosomally integrated transgene". Neither Zhang 1995 or Zhang 1993 teach chromosomal integration of DNA constructs; rather, both references disclose only transient transfection experiments which do not require chromosomal integration before gene expression is measured. As discussed above with reference to the Zhang 1995 reference, both references, in fact, teach away from chromosomal integration because the goal of the authors is to study the effect of sequence elements on gene expression in the absence of confounding position effects that may arise upon chromosomal integration. Secondly, because of this teaching away, one of skill in the art upon reading the Zhang references would not be motivated to modify these references to provide for transfectants in which the DNA constructs are chromosomally integrated.

Thirdly, an additional teaching away from the invention of the present application is found in the Zhang 1993 reference. Figure 7 and its accompanying text at page 2304, second full paragraph after the figure and figure legend, indicates that when the point mutation, TCTGAGTCA (SEQ ID NO:1), is used in a DNA construct for transient transfection studies of expression, the authors found that "the point mutation reduced the expression level by approximately 70%" as compared to the wild type sequence. Such a reduction in gene expression using the point mutation of SEQ ID NO:1 would discourage one of skill in the art from employing this sequence to achieve the expression of target genes.

Finally, a *prima facie* case of obviousness may be rebutted by a showing of superior or unexpected results. MPEP 2144.09. In light of the reduction in the gene expression of GH when the TCTGAGTCA sequence element was used to direct the expression of GH as documented in Zhang 1993, the results of the present inventors as shown *inter alia* in Table 1 of the present application represent an unexpected result. The data in Table 1 shows not a 70% reduction in the expression of GH as reported in Zhang 1993, but rather increases of 13- and 26-fold over wild type (single copy integrated), 18-fold over wild type (10 copies integrated), and 25-fold over wild type (13 copies integrated).

Another unexpected result is the ability of the TCTGAGTCA (SEQ ID NO:1) sequence when used in an expression construct to over come previous limitations associated with expression of chromosomally integrated constructs (e.g., position-effect variegation, silencing of transgenes, and the inability to increase expression by increasing gene copy number). See, e.g., Sabl et al., Genetics 142:447-458 (1996); Palmer et al., Sharpe et al., EMBO J 11:4565-4572 (1992); and Chen et al., Proc. Natl. Acad. Sci. USA 94:5798-5803 (1997). Again, the data in Table 1 shows that these problems have been eliminated through the use of SEQ ID NO:1 in constructs. Table 1 shows that when the mutant HS-40 transgene is used, a strong positive correlation between copy number of the transgene and hGH expression is observed; in contrast, when the wild type HS-40 transgene is used, increased expression of hGH is not consistently observed with increased copy number.

In view of the foregoing arguments, Applicants submit that the Examiner's *prima facie* case of obviousness has been rebutted. Accordingly, Applicants respectfully request that this ground for rejection be withdrawn.

In view of the above, each of the presently pending claims in this application is believed to be in immediate condition for allowance. Accordingly, the Examiner is respectfully requested to withdraw the outstanding rejection of the claims and to pass this application to issue. If it is determined that a telephone conference would expedite the prosecution of this application, the Examiner is invited to telephone the undersigned at the number given below.

In the event the U.S. Patent and Trademark office determines that an extension and/or other relief is required, applicant petitions for any required relief including extensions of time and authorizes the Commissioner to charge the cost of such petitions and/or other fees due in connection with the filing of this document to **Deposit Account No. 03-1952** referencing docket no. 514162000120. However, the Commissioner is not authorized to charge the cost of the issue fee to the Deposit Account.

Dated: August 1, 2005

Respectfully submitted,

By _____

Otis Littlefield
Registration No.: 48,751
MORRISON & FOERSTER LLP
425 Market Street
San Francisco, California 94105-2482
(415) 268-6846